

Interaction of Ambient Temperature and
Microwave Power Density on
Schedule-Controlled Behavior in the Rat

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Interaction of ambient temperature and microwave power density on schedule-controlled behavior in the rat

Michael I. Gage and W. Mark Guyer

Neurotoxicology Division, MD-74B, Health Effects Research Laboratory, U.S. Environmental Protection Agency
Research Triangle Park, North Carolina 27711

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Most guidelines of microwave exposure do not explicitly address effects of ambient temperature. Our experiment revealed that ambient temperature potentiates the behavioral effects of intensity of irradiation. Sixty-four adult male Long-Evans rats were trained to insert their heads into a food cup to obtain food pellets on a 1-min variable-interval schedule of reinforcement. Two groups of four rats each were then exposed to 2450-MHz CW microwaves for 15.5 hours under one of the following eight combinations of power density and air temperature: 8 or 14 mW/cm² at 22°C; 0, 8, or 14 mW/cm² at 26°C; and 0, 8, or 14 mW/cm² at 30°C (relative humidity was 50% in all cases). Response rate of each rat following exposure was compared with its control rate at 0 mW/cm² and 22°C. After exposure at 8 mW/cm², response rates were reduced by a mean of 13.8% at 22°C, 27.5% at 26°C, and 77.5% at 30°C. After exposure at 14 mW/cm², rates were reduced by a mean of 21.1% at 22°C, 43.7% at 26°C, and 80.0% at 30°C. In the absence of microwaves the higher temperatures caused only slight decreases in response rate.

INTRODUCTION

Increases in temperature and humidity can cause an added thermal burden on the human being that can heighten effects of any given exposure to microwaves [Mumford, 1969]. Experimental findings have shown that several types of microwave effects on rodents are enhanced by increasing the ambient temperature during exposure. These findings include decreased survival times of rats [Deichmann *et al.*, 1959]; increased lethality of mice [Rugh *et al.*, 1975]; impaired operant performances of rats [Gage, 1979]; and decreased rates of energy absorption in mice [Monahan and Ho, 1977], which might indicate behavioral activity to minimize microwave absorption or, possibly, heat exhaustion.

Our experiment was designed to quantify more precisely the interactive effects of microwave power density and temperature on behavioral changes measured immediately after termination of exposure. Observations were made on rats of several independent groups under several conditions of exposure. A wider range of temperatures and power densities and a more refined behavioral task were used than were used in prior work [Gage, 1979]. More definite conclusions may therefore be drawn regarding the effects

of temperatures commonly found in the environment on behavioral changes produced by microwaves.

MATERIALS AND METHODS

Subjects

Sixty-four male, experimentally naive Long-Evans rats (obtained from Charles River Breeding Laboratories) were used as subjects. When rats of the first group were exposed to microwaves, they were 288 days old and weighed from 260 to 334 g. Throughout the experiment each rat was maintained at a constant body mass by postsession feeding in its home cage. Water was available during the day except during behavioral testing. All rats were deprived of food and water nightly and were kept in the dark from 1800 to 0600 or 1630 to 0800 during exposures.

Behavior Testing

Behavioral testing took place in a small plastic testing chamber that was housed inside a sound-isolated cubicle. Rats were trained to insert their heads into a food cup at one end of the testing chamber to obtain a 45-mg food pellet on each occasion of reinforcement. Head insertions interrupted an infrared beam of light which activated automatic schedule-programming apparatus located in an adjacent room. Details of this apparatus have been

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presented elsewhere [Gage, 1979].

The rats were tested on a variable-interval 1-min (VI-1) schedule of reinforcement. With this schedule the opportunity to obtain a food pellet occurs on the average of once each minute, according to a preplanned sequence of intervals. This reinforcement opportunity is not signaled. Interval lengths were distributed as a rectangular arithmetic progression, but the sequence of lengths was scrambled [Catania and Reynolds, 1968]. The first head insertion response following a scheduled opportunity for reinforcement produced a food pellet. A reinforcement opportunity was forfeited if no response occurred before a subsequent opportunity for reinforcement was programmed.

Microwave Exposure Apparatus

Exposure to 2450-MHz CW microwaves occurred in an RF anechoic chamber under far-field conditions. Inside the anechoic chamber was a Styrofoam exposure chamber in which ambient air temperature was constant within 1°C and relative humidity was constant within 5% of the stated value. Microwave exposures and exposures to 0 mW/cm² at temperatures other than 22°C were conducted with rats in the Styrofoam chamber. Sham exposures at 22°C were conducted with rats in additional Styrofoam chambers of the same size which had a similar environmental control system. These additional chambers were located near the anechoic chamber or inside a second anechoic chamber.

During all exposures each rat was housed in a small rectangular, visually transparent acrylic box with holes in the sides and slats on the bottom to allow air to flow through the box and excrement to drop below the box. During microwave exposures a group of four rats in boxes was placed inside the Styrofoam chamber in a diamond pattern so the boxes were equidistant from the center on the chamber floor. The rats were approximately 2.7 m below a vertically directed horn antenna. The long axis of the boxes paralleled the magnetic-field vector of the microwave field. During sham exposures, groups of four or eight rats were placed inside one of the other Styrofoam chambers so that rats were equidistant from each other. Further details of the microwave exposure system and the rodent exposure boxes have been described elsewhere [Blackman et al., 1975; Elder and Ali, 1975; Gage, 1979].

Procedure

Behavioral testing sessions occurred daily beginning at 0800. Rats of each group were tested simultaneously in four individual test chambers. The first response in each session produced a pellet. Each session lasted until 30 additional opportunities for reinforcement had been programmed on the VI-1 schedule, which required approximately 30 min. Microwave exposures took place only after at least seven sessions of behavioral testing during which stabilization of responding occurred. During this week the rats became adapted to being in the acrylic exposure boxes overnight and having their rectal temperatures taken.

For three sessions prior to microwave exposure, behavioral testing followed an overnight sham exposure at 22°C. The sham exposures served to acclimate the rats to the housing conditions during exposure. The last sham exposure also served as the 0-mW/cm² at 22°C exposure. One group of four rats was then exposed to microwaves for the 15.5-hour period lasting from 1630 to 0800 hours. Two groups of rats were exposed under each of the following conditions of power density and ambient temperature: 8 or 14 mW/cm² at 22°C; 0, 8, or 14 mW/cm² at 26°C; and 0, 8, or 14 mW/cm² at 30°C. Relative humidity was 50% during all exposures. Air velocity was 0.07 m/s [Smialowicz et al., 1979]. The night following exposure, each group was given a sham exposure at the ambient temperature of the previous night.

Rectal temperature was measured in one rat of each group both before and after each microwave or sham exposure. Behavioral testing occurred approximately 10 min after termination of an irradiation treatment. During this time the rectal temperature was measured, and the rats were transported to the testing room and given access to water. The data were statistically analyzed by analysis of covariance techniques which allowed measures obtained following exposure to be equated with those observed the day before exposure for each rat [Service, 1972].

RESULTS

Control performance of the rats on the VI-1 schedule involved fairly steady responding throughout each session with some oscillations in rate of head insertions over short periods of time during a session. The observed pattern of responding is typical of behavior under variable-interval schedules of reinforcement [Catania and Reynolds, 1968].

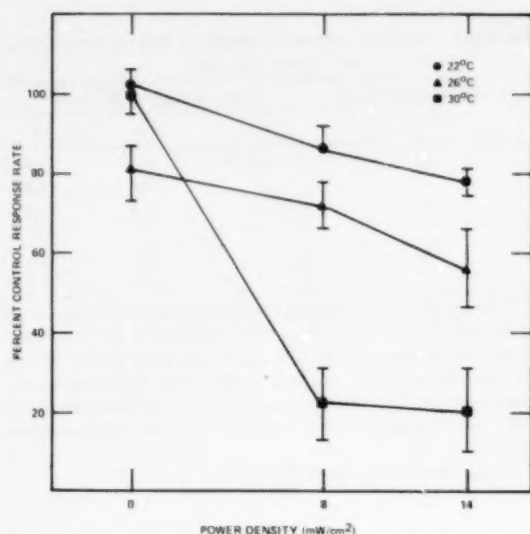


Fig. 1. Effects of power density at three ambient temperatures on response rate calculated for each rat as a percentage of its control response rate of the previous session. Each point represents the mean (plus or minus standard error) of percentage of response rates for all rats treated similarly. Data for the 0-mW/cm², 22°C condition were obtained from 63 rats; data for the 14-mW/cm², 26°C condition were obtained from seven rats; and data for all other points were obtained from eight rats.

Response rates under control conditions differed among the rats. Therefore data were obtained for each rat both before and after microwave exposure so that each rat could be compared in performance with its baseline of behavior.

Effects at 0 mW/cm² and 22°C were evaluated by comparing data obtained on the day prior to microwave exposure with data obtained 2 days prior to microwave exposure. This comparison was possible because testing for 3 days before microwaves followed a night of sham exposure at 22°C for 63 rats. One rat which was scheduled to receive irradiation at 14 mW/cm² and 26°C was excluded from the analysis of this experiment before exposure because of erratic behavioral performance. Effects of the other exposure conditions were evaluated by comparing data following the exposure with data obtained on the day prior to the exposure. Effects of these exposures were based on the data from the eight rats similarly exposed except for the effects under the 14-mW/cm², 26°C condition, which were based on the data of seven rats.

Microwave irradiation reduced rates of responding in direct proportion to the power density at each ambient temperature, as seen in Figure 1. This decrease of rate was most pronounced after exposures at 30°C. Temperature alone did not have a consistent effect. There was a small decrement in response rate after treatment under the 0-mW/cm², 26°C condition but not after that of 0 mW/cm² and 30°C. The mean and standard error of response rates of all the rats after the last sham exposure at 22°C was 0.54 ± 0.04 response/s. Inspection of the cumulative records of responding indicated that the rate was lower throughout sessions following irradiation but that the pattern of fairly consistent responding did not change.

An analysis of covariance confirmed that the effect of power density [$F(2, 54) = 3.4, P < 0.05$] and the interaction of power density and temperature [$F(3, 54) = 4.9, P < 0.005$] on response rates were both significant. The effect of ambient temperature on rate [$F(2, 54) = 7.6, P < 0.002$] was also significant. The analysis showed that the response rate of each rat after exposure was dependent on its control rate [$F(8, 54) = 13.8, P < 0.001$]. During sessions 1 day after microwave exposure many rats showed behavioral overcompensation [$F(1, 54) = 322.3, P < 0.001$]; that is, rates exceeded those measured the day before irradiation. However, there were no significant effects of power density [$F(2, 54) = 1.7$] or of ambient temperature [$F(2, 54) = 0.04$] on response rates during sessions 1 day after irradiation.

In addition to the frequency of head insertions, another parameter of behavior, that of response duration, was measured. The total duration of light beam interruptions was recorded and divided by the number of responses to obtain a mean response duration. The mean response duration of each rat was compared to its mean response duration for the prior day and was analyzed in a manner similar to the response rate measure. The effect of microwave irradiation at each ambient temperature on percent of control mean response duration of the prior session is seen in Figure 2. Increases in response duration occurred after exposures at 14 mW/cm² and 26°C and after exposure at both 8 and 14 mW/cm² when the temperature was 30°C. There were no significant increases in response duration after any exposures at 22°C. The mean response duration of all rats after the last sham exposure at 22°C was 0.81 s (standard error = ± 0.05 s).

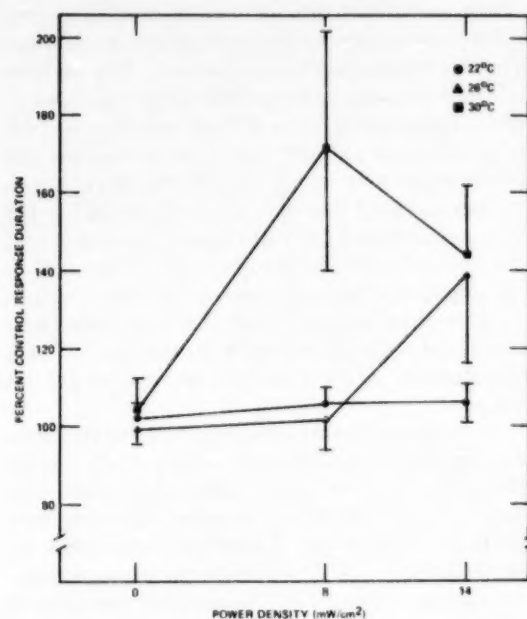


Fig. 2. Effects of power density at three ambient temperatures on mean response duration as calculated for each rat as a percentage of its control value of the previous session. Each point is the mean (plus or minus standard error) of all rats exposed under a stated condition. The number of rats under each condition is the same as stated in Figure 1.

An analysis of covariance showed that the effect of ambient temperature on response duration was significant [$F(2, 54) = 3.9, P < 0.03$], but the effects of power density [$F(2, 54) = 1.4, P = 0.25$] and of the interaction between power density and temperature [$F(3, 54) = 2.0, P = 0.12$] were not significant. The lack of a significant effect of power density on this measure was probably due to the large interanimal variability of durations after exposure to microwaves. Mean response duration of each rat after exposure was dependent on its control performance [$F(8, 54) = 10.0, P < 0.001$]. Overcompensation during sessions 1 day after exposure was also observed [$F(1, 54) = 107.8, P < 0.001$]. As was the case for response rate 1 day after exposure, there were no effects of power density [$F(2, 54) = 1.6$] or of ambient temperature [$F(2, 54) = 0.7$] on means of response duration.

Changes of rectal temperature after microwave exposures differed from those after sham exposures in the same rat. These changes are shown in Table 1 for

TABLE 1. Rectal temperature changes in degrees Celsius after microwave exposures.

Ambient temperature (°C)	Power density (mW/cm ²)		
	0	8	14
22	-1.5 ± 0.24*	-1.7	-0.9
		-1.8	-1.8
26	-1.8	-1.4	-1.3
	-2.0	-1.7	+0.1
30	-1.1	-0.4	0.0
	-2.3	-0.5	+2.7

Data are differences in temperature at 0800 hours from that at 1600 hours of the previous day as measured in the same rats. Shown are the temperature differences measured in each of two rats for each exposure condition. Temperature difference for the 0-mW/cm², 22°C condition was obtained from the same rats in which temperature was also measured after the other exposure conditions.

*Mean and standard error for 16 rats.

the one animal in each group from which temperature data were recorded. In the afternoon, before the sham exposure condition identified in Table 1 as 0 mW/cm² at 22°C, the mean and standard error of the rectal temperature of all rats was $38.24^\circ \pm 0.13^\circ\text{C}$. The temperature of all rats before their exposure on the next afternoon was $38.14^\circ \pm 0.16^\circ\text{C}$. Table 1 shows that body temperature decreased after the sham exposures and after all but the highest microwave exposures at the elevated ambient temperatures. While temperatures of relatively few rats after each exposure condition were measured in this experiment, a trend toward lessened decreases of increased rectal temperature with increasing power densities and ambient temperature was observed.

Small changes in body mass were seen after exposures, as illustrated in Table 2. Mass was increased from that on the prior day with increased ambient temperature in the absence of microwaves and at

TABLE 2. Percentage of body mass (relative to control values) after microwave exposures.

Ambient temperature (°C)	Power density (mW/cm ²)		
	0	8	14
22	99.8 ± 0.02*	101.3 ± 0.31	102.0 ± 0.49
26	101.4 ± 0.27	101.4 ± 0.52	97.6 ± 1.24†
30	101.2 ± 1.09	99.1 ± 0.56	97.8 ± 0.55

Data are the means and standard errors for all the rats exposed under each condition (for eight rats, except as noted) and are expressed as a percentage of the body mass of the prior day.

*For 63 rats.

†For seven rats.

22°C in the presence of microwaves. Mass decreased after exposures to microwaves at the higher ambient temperatures. An analysis of covariance showed that effects of power density [$F(2, 50) = 5.8, P < 0.006$], of ambient temperature [$F(2, 50) = 10.8, P < 0.001$], and of the interaction of power density and temperature [$F(3, 50) = 4.2, P < 0.02$] on body mass were significant. The analysis indicated that body mass after exposure was dependent on each rat's control mass [$F(8, 50) = 50.6, P < 0.001$]. Means of body mass 1 day after exposure were higher than on the day before exposure [$F(1, 50) = 445.8, P < 0.001$]. There were no effects on body mass of power density [$F(2, 50) = 2.9, P = 0.07$] or of ambient temperature [$F(2, 50) = 1.4, P = 0.26$] 1 day after the exposure.

Specific absorption rate (SAR) has previously been reported for rats of similar size exposed four at a time in the same exposure apparatus [Gage, 1979]. Those SAR's were determined by the twin-well calorimetric technique as described by Kinn [1977]. The mean SAR was 0.20 ± 0.06 mW/g per mW/cm² of incident radiation. There were differences arising from position in the exposure chamber that resulted in a range of SAR's from 0.14 ± 0.01 to 0.26 ± 0.04 mW/g. No clear relation between rank ordering of behavioral changes and chamber position was observed.

DISCUSSION

The results of our experiment show that intensity of microwave irradiation and ambient temperature interact to increase decrements in rates of behavioral responding measured at the termination of irradiation. This dependence of the magnitude of response rate decrement in operant, schedule-controlled performance on the ambient temperature during microwave irradiation reaffirms results of earlier behavioral experiments [Gage, 1979].

Increases of body temperature and losses of body mass, although slight and within physiologically normal limits, were observed along with the behavioral changes after combined exposure to higher levels of temperature and microwave irradiation. Increases in rectal temperature after 1 or 2 hours of exposure to microwaves at power densities in the range 1–10 mW/cm² at ambient temperatures of 24°C have been reported by Lu *et al.* [1977]. After long exposures, temperatures reverted to control values except after exposures at 20 mW/cm². The interplay between

temperature changes measured after microwave exposure and circadian rhythmic variation in normal rectal temperature described by Lu *et al.* [1977] was also seen in this experiment, but for a different time of day. The rectal temperature of rats during sham exposures dropped between 1600 and 0800 hours. Less of a temperature decrease over this period might be comparable to a temperature rise during the daylight hours when Lu *et al.* [1977] exposed their rats.

Although the measured changes of temperature and body mass after most microwave exposures were within normal physiological limits (considering their range over a 24-hour period), they may be atypical for a rat at the time of day when measured. Consequently, such changes, which at other times may be normal, may exert abnormal influences on the animal's functional capacities. The behavioral change may reflect or be an index of a change in the overall physiological state of the exposed organism [D'Andrea *et al.*, 1976, 1977]. Contrariwise, the behavioral change can be viewed as a response to the stimulus of the change in some parameters of physiological state [de Lorge, 1976].

The continuum of the microwave-temperature interaction is illustrated by the close agreement of the following data with a prediction. The bisection of the difference between the values for percent of control response rate after exposure at 8 mW/cm² and 26°C and after exposure at 8 mW/cm² and 30°C, shown in Figure 1, predicts that an overnight exposure at 8 mW/cm² under an ambient temperature of 28°C would result in a response rate decrease to 47.5% of the control rate on the prior day. An exposure at 8 mW/cm² and 28°C of a group of four rats exposed previously at 0 mW/cm² and either 26° or 30°C resulted in a mean response rate of $59.7 \pm 12.7\%$ of control rates, and a similar exposure of a different group of four rats exposed previously at 14 mW/cm² and 30°C resulted in a mean response rate of $56.8 \pm 11.8\%$ of control rates.

The effects observed were transient. Twenty-four hours after exposure, all measures returned to baseline levels or even showed overcompensation. None of the measures on the day after exposure were related to the parameters of microwave exposure conditions. Recoverability of baseline has been suggested or reported in other experiments of effects of microwaves on behavior [D'Andrea *et al.*, 1976, 1977; de Lorge, 1976].

The data of our experiment clearly indicate that

ambient temperature is an important factor in determining the potential hazards of microwave irradiation. Conditions of temperature and humidity have long been recognized [Mumford, 1969] as contributing to the possible effects of microwaves. Consequently, it is crucial to specify and maintain conditions of temperature and humidity in laboratory experiments on biological effects of microwaves. Furthermore, it is crucial to examine biological effects of microwaves over the range of temperature and humidity conditions that can be found in the environment if a relation between laboratory findings and real world conditions is sought.

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